



BIOTECON

Diagnostics

For food testing purposes

FOR *IN VITRO* USE ONLY

foodproof[®] Magnetic Preparation Kit III

Version 3, July 2015

For the automated isolation of DNA from raw material and food products of plant and animal origin using the KingFisher[®] Flex instrument

Order No. S 400 13 L

Kit for 5 x 96 isolations

Store the **Magnetic Beads** at 2-8 °C

Store lyophilized **Proteinase K** at 2-8 °C

Store all other kit components at room temperature



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1. Kit Components

Kit Contents

Bottle/Tube/ consumable	Label	Contents / Function
Bottle No.1	foodproof Magnetic Preparation Kit III Lysis Buffer	<ul style="list-style-type: none">• 3 x 250 ml• For lysis of cells and extraction of DNA
Bottle No.2	foodproof Magnetic Preparation Kit III Binding Buffer	<ul style="list-style-type: none">• 2 x 24 ml, add 2 x 56 ml absolute isopropanol• For binding of DNA to the magnetic beads
Bottle No.3	foodproof Magnetic Preparation Kit III Wash Buffer I	<ul style="list-style-type: none">• 3 x 80 ml, add 3 x 80 ml absolute ethanol• For removing impurities
Bottle No.4	foodproof Magnetic Preparation Kit III Wash Buffer II	<ul style="list-style-type: none">• 5 x 60 ml, add 5 x 140 ml absolute ethanol• For removing impurities
Bottle No.5	foodproof Magnetic Preparation Kit III Elution Buffer	<ul style="list-style-type: none">• 110 ml• For elution of DNA
Tube No.6	foodproof Magnetic Preparation Kit III Proteinase K (working solution)	<ul style="list-style-type: none">• 20 x lyophilized• For protein digestion and inactivation of endogenous nucleases.• Dilute Proteinase K by addition of 1.8 ml of ddH₂O, mix thoroughly and store like described below!
Tube No.7	foodproof Magnetic Preparation Kit III Magnetic Beads	<ul style="list-style-type: none">• 10 x 1,1 ml• For binding of DNA
Consumable	Deep Well Plate	<ul style="list-style-type: none">• 5 x 4 plates
Consumable	Elution Plate	<ul style="list-style-type: none">• 5 x 2 plates• Elution Plates and Tip Plates are identically. Use one provided Elution Plate as a Tip Plate.
Consumable	Tip Comb 96 DWH	<ul style="list-style-type: none">• 5 x 1 comb
Consumable	Adhesive Seal	<ul style="list-style-type: none">• 5 x 2 foils



Chemical Hazard

When working with chemicals, always wear a lab smock, disposable gloves and protective goggles. Observe the legal requirements for working with biological material! For more information, please consult the corresponding material safety data sheet (MSDS).

If any reagent bottles are damaged or leaky, wear gloves and protective goggles when discarding the bottles to avoid injuries.

European Community risk and safety phrases for the components of the **foodproof** Magnetic Preparation Kit III, to which they apply, are listed below:

Lysis Buffer



danger
H302-319 P305-351-338

Proteinase K:



danger
H315-319-334-335 P280-305-
351-338-310-405

Binding Buffer



danger
H225-319-336 P210-233-305-351-338

Wash Buffer I



warning
H302-312-332-412 EUH032 P273

H315:	Causes skin irritation.
H319:	Causes serious eye irritation.
H334:	May cause allergy or asthma symptoms or breathing difficulties if inhaled.
H335:	May cause respiratory irritation.
H225:	Highly flammable liquid and vapor.
H336:	May cause drowsiness or dizziness.
H302:	Harmful if swallowed.
H312:	Harmful in contact with skin.
H332:	Harmful if inhaled.
H412:	Harmful to aquatic life with long lasting effects.
EUH032:	Contact with acids liberates very toxic gas.
P280:	Wear protective gloves/protective clothing/eye protection/face protection.
P305+P351+P338:	If in eyes: Rinse cautiously with water for several minutes. Remove contact lenses, if present and easy to do. Continue rinsing.
P310:	Immediately call a POISON CENTER or doctor/physician.
P405:	Store locked up.
P210:	Keep away from heat/sparks/open flames/hot surfaces. — No smoking.
P233:	Keep container tightly closed.
P273:	Avoid release to the environment.



Number of Preparations

5 x 96 isolations

Storage

All buffers and kit components of the **foodproof** Magnetic Preparation Kit III, except Proteinase K and Magnetic Beads, should be stored at room temperature* and are stable for at least 12 months under these conditions.

Proteinase K: Lyophilized Proteinase K should be stored at 2-8 °C. Dissolved Proteinase K must be stored at -20 °C. Avoid multiple freezing and thawing cycles of diluted Proteinase K.

Magnetic Beads: The magnetic particles should be stored at 2-8 °C.

Wash Buffer I and II: Wash Buffers charged with ethanol should be stored at room temperature and must be sealed accordingly. If any precipitates are visible within the provided solutions solve them by carefully heating up to 30 °C.

*room temperature (RT) is defined as range from 15-30 °C.

2. How to Use this Product

2.1 Product Overview

Test Principle

The **foodproof** Magnetic Preparation Kit III in combination with the KingFisher® Flex instrument provides fully automated purification of total genomic DNA from up to 200 mg from raw material and food products of plant and animal origin. The DNA isolation process is based on patented magnetic bead technology, which relies on the interaction of nucleic acids with coated magnetic particles under suitable buffer conditions. The kit provides high-quality DNA, which is suitable for direct use in PCR applications.

The KingFisher® Flex instrument performs all steps of the DNA purification procedure automatically. The procedure requires only minimal interaction by the user, namely the initial loading of the system and the preparation of the sample material. Sample cross-contamination and reagent cross-over is effectively eliminated by the provided purification assay.

The KingFisher® Flex instrument uses magnetic rods to transport the DNA, bound to magnetic particles, through the various purification phases: binding, washing and elution. The volume of buffers and other liquids necessary for DNA isolation is reduced to a minimum. Eliminating the direct liquid handling and increasing the automation level results in a fast, reliable and robust technique.

To achieve an efficient lysis and high DNA yields, the samples are first lysed with optimized lysis buffer and Proteinase K in a separate step. After lysis, the DNA binds to the magnetic beads whereas contaminations, metabolites and enzyme inhibitors are efficiently removed during the following three wash steps. Finally, highly purified DNA is eluted in Elution Buffer.

The purified, high quality DNA is ready to use for following downstream applications like PCR or can be stored at -20°C for subsequent use.

Basic Steps

Step	Description
1	Sample lysis by incubation with the foodproof Magnetic Preparation Kit III Lysis Buffer and foodproof Magnetic Preparation Kit III Proteinase K
2	DNA is bound to magnetic beads
3	Washing of bound DNA to remove proteins, and other cellular impurities
4	Purified DNA is recovered using the Elution Buffer

Sample Material

200 mg sample material from raw material and food products from plant and animal origin are processed with the kit.

Quality Control

Each component of the Magnetic Preparation Kit III has been tested against predetermined specifications to ensure consistent product quality.

3. Procedures and Required Materials

3.1 Before you begin

Preparation of Kit Working Solutions

Before starting a run, bring all reagents to room temperature. Gently mix and redissolve any precipitates by warming up to 30°C. Swirl gently to avoid foaming.

foodproof Magnetic Preparation Kit III Lysis Buffer, Binding Buffer and Elution Buffer are ready-to-use. Add the required amount of ddH₂O to the reaction tube containing the **foodproof** Magnetic Preparation Kit III Proteinase K. Vortex for 5 s and keep the solution on ice. Store unused and diluted **foodproof** Magnetic Preparation Kit III Proteinase K at –20 °C.

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Bottle	Content	Preparation of working solution	Storage and stability
2	foodproof Magnetic Preparation Kit III Binding Buffer	Add 56 ml absolute isopropanol to the bottle Binding Buffer. Mix thoroughly and always keep the bottle firmly closed. Note: Check the box on the label of the bottle after isopropanol has been added. Add the date for verifiability.	Store at room temperature. Stable until the expiry date printed on kit label.
3	foodproof Magnetic Preparation Kit III Wash Buffer I	Add 80 ml absolute ethanol to the bottle Wash Buffer I. Mix thoroughly and always keep the bottle firmly closed. Note: Check the box on the label of the bottle after ethanol has been added. Add the date for verifiability.	Store at room temperature. Stable until the expiry date printed on kit label.
4	foodproof Magnetic Preparation Kit III Wash Buffer II	Add 140 ml absolute ethanol to the bottle Wash Buffer II. Mix thoroughly and always keep the bottle firmly closed. Note: Check the box on the label of the bottle after ethanol has been added. Add the date for verifiability.	Store at room temperature. Stable until the expiry date printed on kit label.
6	foodproof Magnetic Preparation Kit III Proteinase K	Dissolve Proteinase K by addition of 1.8 ml double-distilled water (ddH ₂ O) per reaction tube needed.	Store at -20 °C, Stable for 12 months.

Additional Equipment and Reagents required

- KingFisher® Flex instrument
- Measuring cylinder (250 ml)
- Pipette and pipette tips
- Disposable gloves
- ddH₂O
- Vortexer
- absolute Ethanol (96-98 %)
- absolute Isopropanol (96-98 %)
- 2 ml reaction tubes
- Thermomixer for 2 ml reaction tubes (e.g. Eppendorf ThermoMixer® comfort)



3.2 Protocol

Caution

Always wear gloves during the procedure, and follow safety precautions to minimize contact when handling. Follow generally applicable safety precautions regulating the work with biohazard materials. Properly dispose of all contaminated materials, decontaminate work surfaces, and use a biosafety cabinet whenever aerosols might be generated.

The following protocol describes the automated DNA isolation from 200 mg sample material with the KingFisher® Flex instrument:

- Transfer up to 200 mg homogenized sample material into a 2 ml reaction tube and add 1400 µl of **Lysis Buffer** and 70 µl of **Proteinase K**.
- Incubate for 30 min at 65 °C with shaking in a thermomixer.
- Centrifugate for 10 min at 12,000 g.
- Switch on the KingFisher® Flex instrument.

Note: *Before starting the purification process with the KingFisher® Flex instrument please read carefully the user manual! Resuspend/Vortex the Magnetic Beads thoroughly directly before use!*

- **Tip Plate:** Place the Tip Comb 96 DWH on a Tip Plate (Use one provided Elution Plate as Tip Plate. These are identical.).

Prefill the Binding Plate, the Washing Plates and the Elution Plate as described below:

- **Binding Plate:** Add 250 µl **Binding Buffer** and 20 µl **Magnetic Beads**
- **Washing Plate I:** Add 600 µl **Wash Buffer I**
- **Washing Plate II:** Add 800 µl **Wash Buffer II**
- **Washing Plate III:** Add 800 µl **Wash Buffer II**
- **Elution Plate:** Add 200 µl **Elution Buffer**
- Transfer 500 µl of the lysate into the **Binding Plate**.
- Choose assay file "**foodproof_MPK_III**" on instrument and press "START".
- Follow instructions on the instruments display and load the prefilled buffer plates in the right position. Confirm with "START" after each loading step, the instrument then will provide the next free loading position automatically.
- When all plates are loaded, press "START" again to initialize the program.



The following purification steps will run automatically on the KingFisher® Flex System:

- **Binding of the DNA:** Automatically sample mixing for 5 min. Magnetic Beads separation. Transfer of the magnetic particles to Washing Plate I.
- **First Washing:** Automatically sample mixing for 1.5 min. Magnetic Beads separation. Transfer of the magnetic particles to Washing Plate II.
- **Second Washing:** Automatically sample mixing for 1 min. Magnetic Beads separation. Transfer of the magnetic particles to Washing Plate III.
- **Third Washing:** Automatically sample mixing for 1 min. Magnetic Beads separation.
- **Drying:** Drying of the Magnetic Beads outside Washing Plate III for 5 minutes. Transfer of the Magnetic Beads to the Elution Plate.
- **Elution of the DNA:** Incubation of magnetic particles in the Elution Buffer for 10 minutes at 90°C by continuously mixing. Magnetic Beads separation. The Magnetic Beads will automatically be removed and transferred in Washing Plate III (disposal).

Note:

1. *After finishing the extraction protocol, the Elution Plate contains the extracted DNA.*
2. *If the extracted DNA contains carryover of magnetic particles, transfer the DNA into a 1.5 ml reaction tube and centrifuge at maximum speed for 1 minute. Transfer the clear supernatant (contains the DNA) into a new tube.*
3. *Storage of Samples:*

If you want to...	Then
Continue	Use the eluted DNA directly
Stop	The eluted DNA can be stored for several weeks at 4-8 °C or stored at – 20 °C for long-term storage.



3.3 Self-programming of the KingFisher® Flex instrument

Protocol information

Protocol name: foodproof_MPK_III

Kit name: foodproof MPK III

Description: KingFisher® Flex protocol for isolation of genomic DNA from raw material and food products of plant and animal origin.

Plate layouts

Binding Plate		Microtiter 96 Deep Well Plate	
Name	Well volume [µl]	Total reagent volume [µl]	Type
Sample	500	-	Sample
Binding Buffer	250	-	Reagent
Magnetic Beads	20	-	Reagent
Washing Plate I		Microtiter 96 Deep Well Plate	
Name	Well volume [µl]	Total reagent volume [µl]	Type
Wash Buffer I	600	-	Reagent
Washing Plate II		Microtiter 96 Deep Well Plate	
Name	Well volume [µl]	Total reagent volume [µl]	Type
Wash Buffer II	800	-	Reagent
Washing Plate III		Microtiter 96 Deep Well Plate	
Name	Well volume [µl]	Total reagent volume [µl]	Type
Wash Buffer II	800	-	Reagent
Elution Plate		Microtiter 96 Well Plate	
Name	Well volume [µl]	Total reagent volume [µl]	Type
Elution Buffer	150	-	Reagent
Tip Plate		Microtiter 96 Well Plate	
Name	Well volume [µl]	Total reagent volume [µl]	Type
-	-	-	-



File Steps



Tip 1

Tip Comb 96 DWH



Pick-Up

Tip Plate



Binding

Binding Plate

Beginning of step

Precollect

No

Release time, speed

00:00:10, Fast

Mixing / heating:

Mixing time, speed

00:05:00, Medium

Heating during mixing

No

End of step

Postmix

No

Collect count

5

Collect time [s]

10



Washing 1

Washing Plate I

Beginning of step

Precollect

No

Release time, speed

00:00:10, Fast

Mixing / heating:

Mixing time, speed

00:01:30, Medium

Heating during mixing

No

End of step

Postmix

No

Collect count

4

Collect time [s]

3



Washing 2

Washing Plate II

Beginning of step

Precollect

No

Release time, speed

00:00:10, Fast

Mixing / heating:

Mixing time, speed

00:01:00, Medium

Heating during mixing

No

End of step

Postmix

No

Collect count

4

Collect time [s]

3



Washing 3

Washing Plate III

Beginning of step

Precollect

No

Release time, speed

00:00:10, Fast

Mixing / heating:

Mixing time, speed

00:01:00, Medium

Heating during mixing

No

End of step

Postmix

No

Collect count

4

Collect time [s]

3



Dry1

Washing Plate III




Dry time

00:05:00

Tip position

Outside well / tube



	Eluting	Elution Plate	
	Beginning of step	Precollect	No
	Mixing / heating:	Release time, speed	00:00:10, Medium
		Mixing time, speed	00:10:00, Medium
		Heating temperature [°C]	90
	End of step	Preheat	No
		Postmix	No
	ReleaseBeads1	Collect count	4
		Collect time [s]	3
	Leave	Washing Plate III	
		Release time, speed	00:00:10, Fast
		Tip Plate	

4. Typical Results

4.1 Purity

Purified DNA is free of other cellular components and DNA polymerase inhibitors.



5. Troubleshooting

Problem	Possible Cause	Recommendation
Low DNA yield or purity	Kit stored under non optimal conditions.	<ul style="list-style-type: none">• Store the Magnetic Beads at 2-8 °C• Store lyophilized Proteinase K at 2-8 °C• Store diluted Proteinase K at -20 °C• Store all other kit components at room temperature!
	Buffer or other reagents were exposed to conditions that reduced their effectiveness.	<ul style="list-style-type: none">• Store all buffers at room temperature• Close all reagent bottles tightly after each use to preserve pH and stability, and to prevent contamination• After lyophilized reagents are reconstituted, store at -20 °C
	Ethanol not added to Wash Buffer I or Wash Buffer II.	<ul style="list-style-type: none">• Add absolute ethanol to the Wash Buffer I and Wash Buffer II before using• After adding ethanol, mix the Wash Buffer I and Wash Buffer II well, and store at room temperature• Always mark the Wash Buffer I and Wash Buffer II bottle to indicate the addition of ethanol
	Low amount of Magnetic Beads	<ul style="list-style-type: none">• Mix the Magnetic Beads thoroughly before pipetting to the Binding Plate
DNA does not perform well in real-time PCR	Salt carryover during elution	<ul style="list-style-type: none">• Check the Wash Buffers for salt precipitates. If there are any precipitates, solve these precipitates by careful warming• Ensure that Wash Buffers are stored at room temperature
Low A260:A280 ratio from UV measurement, eluted DNA is brown colored	Small part of the magnetic particles are left in the elution	<ul style="list-style-type: none">• Centrifuge at full speed for 1 min and transfer supernatant (contains DNA) to a new tube

6. Appendix

KingFisher® Flex Software

The KingFisher® Flex Software was used to create assay files for the KingFisher® Flex instrument. The respective assay file can either be transferred onto the KingFisher® Flex workstation or be started directly from within the BindIt software. Keep in mind that assay files run directly will not be stored in the workstation memory!



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7. Warranty and Disclaimer of Liability

Limited Warranty and Disclaimer of Liability. BIOTECON Diagnostics warrants that this product is free from defects in materials and workmanship through the expiration date printed on the label and only if the following conditions are complied with:

- (1) The product is used according to the guidelines and instructions set forth in the product literature;
- (2) BIOTECON Diagnostics does not warrant its product against any defects when: defect is a result of material or workmanship not provided by BIOTECON Diagnostics; defect is caused by misuse or use contrary to the instructions supplied; the product is contaminated by improper storage or handling;
- (3) All warranties of merchantability and suitability for a particular purpose, written, oral, expressed or implied, shall extend only for a period of one year from the date of manufacture. There are no other warranties that extend beyond the conditions described here;
- (4) BIOTECON Diagnostics does not assume responsibility to any purchaser of its product for any undertaking, representation or warranty made by any dealers or distributors selling its products beyond those herein expressed, unless deviating terms are expressed in writing by an officer of BIOTECON Diagnostics;
- (5) BIOTECON Diagnostics does not assume responsibility for incidental or consequential damages, including, but not limited to responsibility for loss of use of this product, removal or replacement labor, loss of time, inconvenience, expense for telephone calls, shipping expenses, loss or damage to property or loss of revenue, personal injuries or wrongful death;
- (6) BIOTECON Diagnostics reserves the right to replace or allow credit for any modules returned under this warranty.

8. Supplementary Notes

8.1 Ordering Information

Order No.: S 400 13 L

Please send your order directly to order@bc-diagnostics.com or call us at +49 (0)331-2300-200.

BIOTECON Diagnostics offers a broad range of reagents and services. For a complete overview and for more information, please visit our website at www.bc-diagnostics.com.

8.2 Trademarks

foodproof® is a trademark of BIOTECON Diagnostics GmbH.

KingFisher® is a registered trademark of Thermo Fisher Scientific.

Other brand or product names are trademarks of their respective holders.

8.3 Contact and Support

If you have questions about this or any other product of BIOTECON Diagnostics, please contact our Technical Support, contact details at www.bc-diagnostics.com. Our expert scientists will provide any request with rapid and effective help. We also welcome you to contact us if you have suggestions for enhancing our product performance or using our products in new or specialized ways.



9. Change Index

Version 1, August 2013

First version of the package insert.

Version 2, April 2014

Page 4: Change in volume of Proteinase K and double-distilled water (ddH₂O) added

Page 8: Change in volume double-distilled water (ddH₂O) added

Page 9: Change in volume of Proteinase K added

Version 3, July 2015

Page 3: Change in volume double-distilled water (ddH₂O) added

Page 7: Change in volume double-distilled water (ddH₂O) added

Page 8: Change in volume of Proteinase K added

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